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ETHYLENE DIBROMIDE FOR CONTROL OF THE EUROPEAN CHAFER IN SOILS ACCOMPANYING NURSERY PLANTS

Agricultural Research Service
United States Department of Agriculture

ETHYLENE DIBROMIDE FOR CONTROL OF THE EUROPEAN CHAFER IN SOILS ACCOMPANYING NURSERY PLANTS[1/]

By H. Tashiro, Entomology Research Division[2/]

The use of ethylene dibromide to eliminate the European chafer (Amphimallon majalis (Raz.)) from the soil of nursery plants is authorized by the Department of Agriculture (5) to satisfy the requirements of the quarantine against this insect. Several methods of application are approved. This paper describes studies conducted during 1956-57 that resulted in the approval of the following methods of applying ethylene dibromide: (1) Immersing balled and burlapped and potted soil in dilute aqueous solutions, (2) pouring dilute aqueous solutions on the surface of the soil of potted plants, and (3) injecting more concentrated forms into the soil of balled and burlapped and potted plants.

The earlier work by Gambrell and Mason (2) in investigating the insecticidal and phytotoxic properties of this fumigant and the studies by Mason and Chisholm (3) in determining its effectiveness against the Japanese beetle (Popillia japonica Newm.) served as guides to the immersion and pour-on tests. The work of Fleming et al. (1) served as a guide to the injection tests. Their method eliminates two disadvantages of the immersion and pour-on treatments: The need for a large quantity of water and the water-logged condition of the treated soil.

These treatments must be safe for nearly all nursery-grown plants to be practical. The reaction of 16 species of balled and burlapped evergreens, 28 species of flowers and tropical foliage plants, and 17 varieties of vegetable transplants to ethylene dibromide was determined. The work was done in cooperation with four nurserymen and seven florists located in the European chafer-infested areas of western New York.

1/ In cooperation with the Department of Entomology, New York State Agricultural Experiment Station, Geneva.

2/ Assisted by E. L. Tuttle (resigned), G. R. Fryer, and I. D. Smith (resigned) of the Entomology Research Division.

FORMULATIONS OF ETHYLENE DIBROMIDE USED[3/]

Five formulations, prepared by chemists of the Entomology Research Division, were used during the course of these studies. All were miscible in nearly all proportions with water and formed solutions that were almost clear. Three of these formulations were used for the immersion and pour-on treatments, two for the injection treatments.

Immersion and Pour-On

Work was initiated in 1956 with formulation 253, then authorized for use against the Japanese beetle. Its composition is as follows:

	Percent by weight
Ethylene dibromide	13.0
Chlordane (technical)	6.5
Cellosolve (ethylene glycol monoethyl ether)	6.5
Tween 20 (polyoxyalkylene derivative of sorbitan monolaurate)	6.5
Isopropyl alcohol (99%)	67.5

A formulation temporarily known as IDM-401, without chlordane, was compared with formulation 253. Its composition is as follows:

	Percent by weight
Ethylene dibromide	19.5
Tween 20	6.5
Isopropyl alcohol (99%)	74.0

It became necessary early in these studies to develop a new formulation containing Dowfume W-85 (83% ethylene dibromide and 17% light petroleum by weight) because of patent restrictions on the use of ethylene dibromide. Formulation 253 was replaced with formulation 431, which is presently approved. Its composition is as follows:

	Percent by weight
Dowfume W-85	24.1
Chlordane (technical)	10.0
Cellosolve	10.0
Tween 20	20.0
Isopropyl alcohol (99%)	35.9

3/ The mention of a trade product does not imply its endorsement by the U.S. Department of Agriculture over similar products not named.

Injection

Much of the preliminary work was performed with a miscible ethylene dibromide formulation of the following composition:

	Percent by weight
Ethylene dibromide	2.5
Tween 20	2.5
Isopropyl alcohol (99%)	95.0

When it became necessary to replace technical ethylene dibromide with Dowfume W-85, a new formulation, hereafter referred to as formulation 434, was developed. It contains the following ingredients:

	Percent by weight
Dowfume W-85	3.1
Tween 20	2.9
Isopropyl alcohol (99%)	94.0

TESTS AGAINST THIRD-INSTAR GRUBS

Most of the studies dealt with third-instar grubs. This stage has the longest duration in the life cycle, extending from September through May. It presents the greatest hazard of spreading infestations through the movement of soil.

General Procedures

Preparation of Infested Soils

Both natural and artificially prepared soil balls and soil in pots were treated. Spirea plants growing in Ontario loam were dug in the fall with soil balls 6, 8, and 10 inches in diameter and equivalent depths, wrapped in burlap, and held in storage until needed. The fibrous roots of the plants provided food for introduced grubs. Artificial soil balls were prepared by compressing friable Farmington loam into cylinders of hardware cloth 8 inches in diameter and 8-10 inches deep and lined with burlap. Clover and grass seed was distributed in the soil to provide young sprouts as food for the grubs. Wheat was planted in standard 4-inch clay pots and maintained until a solid mass of roots had developed along the inside of the pots. A screen placed in the bottom of each pot prevented the escape of introduced grubs through the drainage hole.

The soils were infested with third-instar grubs a week before treating to permit them to adjust to their new environment. Five individuals were introduced into each pot and 25 into each burlapped soil ball by placing them on the soil surface and allowing them to burrow in.

Treatment of Soils with Fumigant

For the immersion treatments each ball or pot was submerged separately in water containing the fumigant until the soil became saturated, then removed from the treating solution, and permitted to drain. Saturation was assured when bubbling ceased. Checks were immersed in water only until saturated.

Pour-on treatments were applied only to potted soil; an aqueous solution of the fumigant was applied to the surface. A volume equivalent to either one-fifth or one-tenth of the soil volume in the respective pots was added. Check pots received the same amount of water only.

Ethylene dibromide was injected into the soil balls and pots with the same modified veterinary hypodermic syringe used by Fleming et al. (1) In injecting soil balls more than 6 inches in depth, the fumigant was discharged at a depth of 3 inches, but in smaller balls the fumigant was discharged at half the depth of the ball. Injections were spaced evenly over the tops or bottoms of the soil balls when two or more were deemed necessary. Checks received no treatment other than being held with the treated soils.

After treatment the soils were held undisturbed for a week. At the end of this week, hereafter referred to as the exposure period, the grubs were transferred to untreated soil and held at 60 to 70° F. for further observations. They were examined at 3- to 4-day intervals for 2 weeks and at weekly intervals thereafter. Recovery of both treated and untreated grubs was considered very satisfactory; in most tests more than 90 percent of the original numbers introduced were recovered.

Immersion of Soils

Effect of Concentration

Immersion of balled and burlapped nursery stock in a solution containing 3.0 ml. of formulation 253, or 0.36 gram of ethylene dibromide, per gallon of water was the approved treatment against the Japanese beetle. Preliminary tests at this dosage showed that it was inadequate for killing all the chafer grubs in soil balls immersed in it, but 0.72 gram appeared excessive. Soil balls of spirea were immersed in solutions containing four levels of formulation 253 and maintained at an average temperature of 36° F. during the 1-week exposure period. The results given in table 1 show that 0.42 gram of ethylene dibromide per gallon did not kill all the grubs within 5 weeks, but 0.48 gram did so in 3 weeks. A concentration of 0.54 gram per gallon was chosen for further studies of the immersion method of application.

Since a preliminary study indicated that ethylene dibromide might be more effective against chafer grubs in potted soil than in balled and burlapped soil, Farmington loam and Canadian peat in pots were immersed in a series of concentrations of formulation 431 to supply from 0.26 to 0.54 gram of ethylene dibromide per gallon of water

and held at 40° F. during the exposure period. A concentration of 0.4 gram per gallon eliminated all the grubs (table 1) within 5 weeks. Insecticidal action was essentially the same in the two types of soils.

Influence of Formulation

Formulations IDM-401 and 253, ethylene dibromide alone and with chlordane, were compared at a dilution of 0.54 gram of ethylene dibromide per gallon of water to determine the effect of chlordane in the formulation. Formulation 253 introduced 0.27 gram of chlordane per gallon. When the soil of spirea plants was immersed in both formulations and held during the exposure period at different temperatures, as shown in table 2, both formulations gave comparable results. Chlordane had no effect under these conditions. The temperature of 36° F. appeared to be the minimum at which the ethylene dibromide dosage used could effect complete destruction of the grubs.

When it became necessary to substitute Dowfume W-85 for ethylene dibromide, formulations 253 and 431, both containing chlordane, were compared at temperatures of 40, 50, 60, and 70° F. during the 1-week exposure period by immersing soil balls of spirea. Results are shown in table 3. Both formulations were equally effective. Mortality increased progressively with increments of temperature during the exposure period but the effect was not evident during the subsequent holding periods.

Effect of Duration of Immersion

The soils associated with balled and burlapped plants are never consistent in their permeability to water because of variations in texture, compactness, and organic matter. Therefore, the rate of penetration of liquids into and wetting of the soil vary considerably. To determine the need for thorough wetting, spirea balls were immersed for 10 seconds, a period usually not sufficient to produce saturation, and also until saturated in water containing 0.54 gram of ethylene dibromide per gallon. Complete saturation was obtained within 2 to 3 minutes. The plant balls were held at a mean temperature of 36° F. for 1 week after immersion. Results are given in table 4. Some grubs survived immersion for 10 seconds but none survived in thoroughly saturated soil at the end of 3 weeks.

Influence of Soil Type and Temperature

Certain plants, such as azalea and hydrangea, are grown in soils of high organic content or in peat. Under some circumstances the insecticidal action of ethylene dibromide is retarded by such soils (1). Temperature is also an important factor influencing the effectiveness of ethylene dibromide.

Artificially prepared soil balls of Farmington loam and Canadian peat were immersed in water containing 0.54 gram of ethylene dibromide per gallon. After removal and drainage, the balls were held at 32, 45, 60, and 70° F. for 1 week.

Comparable results were obtained with both soils (table 5). In these tests the Canadian peat did not retard insecticidal action, as has been reported for other soils of high organic matter content. (In tests of other application methods, a retarding effect was evident.) Again it was found that this treatment was not completely effective at temperatures in the low 30's.

Potted Farmington loam and Canadian peat were immersed in water containing 0.4 gram of ethylene dibromide per gallon. After removal and drainage, the potted soils were held at temperatures within the range of from 34 to 70° F. for 1 week. The results, also summarized in table 5, were similar to those obtained with balled and burlapped soils, except at the lowest temperature. At 34° F. no retardation of insecticidal activity occurred as it did when balled and burlapped soils were immersed at 32° F.

Pour-On Application to Soils

Effect of Concentration

A test was conducted to determine the dosage required to eliminate the grubs when the fumigant was poured onto the soil surface. In volumes equivalent to one-fifth those of the soil, aqueous solutions of formulations 253 and IDM-401 were applied to the soil surfaces of the pots. These solutions provided 0.4, 0.6, and 0.8 gram of ethylene dibromide per cubic foot of soil and, with formulation 253, 0.2, 0.3, and 0.4 gram of chlordane, respectively. All pots were held between 50 and 55° F. for the 1-week exposure period. The results, summarized in table 6, showed that a dosage of 0.6 gram of ethylene dibromide was inadequate for eliminating all grubs. Some individuals appeared normal even after 5 weeks. A dosage of 0.8 gram killed all the grubs within 4 weeks and was the level chosen for further studies. As with the immersion method of treatment, chlordane did not contribute noticeably to the mortality.

Influence of Soil Type and Temperature

The variability of potting soil and the variations of temperature at which plants are treated and held made it desirable to determine the influence of these two factors on the insecticidal activity of ethylene dibromide as a pour-on treatment. Three soils--Farmington loam, Dunkirk fine sand, and Canadian peat--in pots were treated with ethylene dibromide, at the rate of 0.8 gram per cubic foot, applied in a volume of water equivalent to one-fifth that of the soil. Pots were held during the 1-week exposure period at 40, 50, 60, and 70° F. The results of two tests--with formulations 253 and 531--were nearly the same and are summarized in table 7.

Mortalities increased with increments in temperature during the exposure period. Temperature had no influence thereafter. All the grubs in the mineral soils were killed within 2 weeks, but more than 5 weeks was required to eliminate the grubs in Canadian peat.

Influence of Volume of Treating Solution

A volume of treating solution equivalent to one-fifth the soil volume is the approximate amount that a friable mineral soil will hold with little or no percolation out of the pot. This is the ideal volume to add. It assures the penetration of the fumigant throughout the soil. However, adding this amount to each pot is seldom possible at one time because of the high level of the soil. It must be added gradually. Generally, it is possible to add a volume equivalent to one-tenth that of the soil at one time.

In volumes of insecticidal solution equivalent to one-tenth and one-fifth those of the soil, ethylene dibromide was applied to pots at the rate of 0.8 gram per cubic foot. After treatment the pots were maintained at temperatures from below 40 to 70° F. during the 1-week exposure period.

The results, summarized in table 8, show that the volume of the solution did not influence insecticidal action in Farmington loam. Canadian peat definitely retarded insecticidal activity; in addition, the rate of kill in this peat was slightly but rather consistently less with the smaller volume of treating solution. It was evident that the dosage would have to be increased to kill all the grubs in Canadian peat.

Control in Potted Roses

Many nurseries pot roses in asphalt paper pots. A few roses potted in 7-inch asphalt pots with a 60-40 mixture of mineral soil and peat were obtained from a commercial grower for treating with a surface application of ethylene dibromide. Each pot was infested with 25 third-instar grubs and held for a week to allow the grubs to become distributed throughout the soil mass.

The pots were treated at the rate of 0.8 gram of ethylene dibromide per cubic foot of soil. Half of these were held between 40 and 50° F. and the other half between 35 and 40° F. Check pots receiving water only were held with each group. Grubs were handled in the usual manner after the 1-week exposure period.

The results of two tests are summarized in table 9. With temperatures of 40 to 50° F. during the exposure period, all grubs were killed within 2 weeks in one test and within 5 weeks in the other. At temperatures below 40° F. more than 5 weeks was required to eliminate all the grubs.

Injection of Soils

Effect of Concentration

Injection of miscible ethylene dibromide into balled and burlapped soil at the rate of 0.8 gram per cubic foot is approved by the Plant Pest Control Division (4) as a certification treatment for the Japanese beetle. Injection of the fumigant at the same rate into soil balls infested with European chafer grubs and followed by a 1-week

exposure period at 36 to 44° F. did not appear sufficiently effective against this species. All the grubs were moribund after 2 weeks, but some lingered up to 9 weeks before finally dying. Six-inch soil balls of spirea were injected at rates of 0.8, 1.2, and 1.6 grams of ethylene dibromide per cubic foot. They were held during the exposure period at 36° F.

Grubs were killed only slightly more rapidly at the higher dosages, as shown in table 10. When phytotoxic hazards and the ultimate death of all grubs exposed to these concentrations of the fumigant were considered, the use of a dosage higher than 0.8 gram seemed unwarranted. This level was selected as the standard dosage for conducting other injection studies.

Fleming et al. (1) determined that the dosage of ethylene dibromide injected into potted soil to kill Japanese beetle grubs could be reduced by 50 percent over that required in balled and burlapped soil. Potted Farmington loam and Canadian peat were injected at rates of 0.4, 0.6, and 0.8 gram of ethylene dibromide per cubic foot to determine the dosage necessary to kill chafer grubs. Formulation 434 was diluted with water to introduce the required amount of the fumigant into each pot in 1 or 2 ml. of the solution. Pots were maintained at 65-70° F. during the 1-week exposure period.

Increasing the dosage increased the rate of kill in both soils but this rate was definitely slower in peat, as shown in table 10. A 0.4-gram dosage eliminated the grubs within 6 weeks in mineral soil. Although the grubs in the treated peat were affected within 3 weeks, they did not all die until after 6 weeks. This dosage was selected for further injection studies in potted soils.

Effect of Soil-Ball Size

Six-, 8-, and 10-inch soil balls of spirea were injected at the rate of 0.8 gram of ethylene dibromide per cubic foot. The 6- and 8-inch balls received the injections from the tops of the balls only; the 10-inch balls were injected from both the tops and bottoms. Since 4 ml. was the maximum volume of fumigant discharged at any one insertion of the needle, the numbers of injections varied with the size of the balls. The balls were held at 36-37° F. during the 1-week exposure period. As shown in table 11, the size of the ball had no influence on the insecticidal action of ethylene dibromide.

Influence of Formulation and Soil Temperature

Six-inch soil balls of spirea plants were injected at the rate of 0.8 gram of fumigant per cubic foot with the 2.5-percent miscible ethylene dibromide formulation. Soon after starting these studies it became necessary to substitute Dowfume W-85 for the earlier formulation used. The test was repeated with formulation 434. In both tests soil balls being held at 40, 50, 60, and 70° F. were treated and maintained at these temperatures during the 1-week exposure period.

Differences in formulation did not influence the effectiveness of ethylene dibromide, as shown in table 12. Also, as in the immersion- and surface-application studies, mortality of grubs was influenced by temperature only during the 1-week exposure period.

Influence of Soil Type and Temperature

Organic soils are sometimes known to retard the insecticidal action of ethylene dibromide. Artificial soil balls 6 inches in diameter were prepared with Farmington loam and Canadian peat and infested. They were held at 32, 40, 50, 60, and 70° F. Then they were injected with the fumigant at the rate of 0.8 gram per cubic foot and maintained at these temperatures during the 1-week exposure period.

As shown in table 13, a definite retardation of mortality occurred in the peat as compared with the Farmington loam. Insecticidal action was the slowest in peat at 70° F., in which up to 9 weeks was required to kill all the grubs. Temperature had a definite influence on the insecticidal action in both soils only during the exposure period, with the rate of mortality increasing with each increment in temperature.

Potted Farmington loam and Canadian peat, infested with grubs and held at 33, 40, 50, 60, and 70° F., were injected at the rate of 0.4 gram of ethylene dibromide per cubic foot. Then they were held at the above temperatures for a 1-week exposure period.

Again, a noticeable retardation of insecticidal action occurred in peat, as shown in table 13. Even though all the grubs were moribund within 3 weeks, they did not all die until after 6 weeks. As in the balled and burlapped soil, the treatment in peat at 70° F. produced the slowest rate of kill. As in the other studies, temperature influenced mortality only during the 1-week exposure period. Each increment in temperature produced a higher rate of kill, but this relationship was not observed after the first week.

Influence of Freezing of the Soil

Fleming et al. (1) did not find that freezing of the soil balls was detrimental to the action of ethylene dibromide in killing Japanese beetle grubs. Eight-inch soil balls of Farmington loam, infested with chafer grubs and held at 32 to 34° F., were injected at the rate of 0.8 gram of ethylene dibromide per cubic foot. Half the soil balls were permitted to freeze gradually during the next 3 days. A minimum temperature of 23° F. was reached. The soil balls were held at this temperature until the sixth day after injection, after which they were permitted to thaw gradually. The grubs were removed the seventh day. They had migrated to the center of the balls. Grubs in the unfrozen balls were evenly distributed throughout the soil.

Although many of the grubs died in the untreated balls permitted to freeze, as shown in table 14, those that survived were normal in every respect. All the grubs in the treated balls, whether from frozen balls or not, were killed by the fumigant. Freezing apparently did not interfere with the effectiveness of the treatment.

TESTS AGAINST OTHER STAGES

Limited studies were conducted to determine the susceptibility of eggs, first-instar grubs, pupae, and adults of the chafer to ethylene dibromide. Balled and burlapped spirea with 6-inch balls of Ontario loam, 6-inch artificial soil balls of Farmington loam prepared in the same manner as for tests against third-instar grubs, and 4-inch pots filled with Farmington loam were infested with the various stages. The fumigant was applied by three methods: (1) Immersion of the soil in water containing 0.54 gram per gallon, (2) application of an aqueous solution to the surface of the soil in 4-inch pots so as to provide 0.8 gram per cubic foot, and (3) injection into the soil balls at the rate of 0.8 gram per cubic foot. To serve as checks for the immersion tests, soils were held in water only, and for the pour-on tests amounts of water equal to those used in the treated pots were poured on soil surfaces. The checks for the injection tests received no treatment. During the exposure period the soils were held undisturbed at temperatures ranging from 60 to 80° F. At the end of this period the stages being tested were transferred to untreated soil for further observation. Eggs were examined every 2 or 3 days or until they hatched or decomposed. Other stages were examined at half-week intervals.

Immersion of Soils

Susceptibility of eggs was determined by placing 50 eggs with visible embryos in a cheesecloth bag with soil, enclosing each bag with window screen to prevent crushing the eggs, and placing one bag in the center of each artificial soil ball. The soil balls were treated immediately after preparation and held for 3 days before eggs were recovered.

In treating first-instar grubs, each pot originally infested with 50 eggs was held until most of the eggs had hatched. Pots were then treated and held for 1 week. Due to the rapid death and decomposition of the small grubs, only two were recovered from the treated pots.

Susceptibility of pupae was determined by infesting balled and burlapped spirea with 25 third-instar grubs and holding them until most of the grubs had pupated. Balls were then treated in water containing 0.27, 0.41, and 1.08 gram of ethylene dibromide per gallon in addition to the standard concentration of 0.54 gram per gallon. These were held for 1 week before pupae were recovered.

The results given in table 15 show that the treatments were effective in killing all the eggs (unfortunately the mortality of untreated eggs appeared unusually high), all the first-instar grubs, and all the pupae exposed to all but the lowest concentration.

The few pupae that were alive after 1 week were dead within $1\frac{1}{2}$ weeks. Adults in artificial soil balls were also treated, but because of the high mortality in the checks the results were inconclusive.

Pour-On Application to Soils

Susceptibility of eggs and first-instar grubs was determined by placing 50 eggs with visible embryos in each pot of soil. Pots were treated at once to measure the effect on eggs, and after the eggs hatched, to measure the effect on first-instar grubs. Eggs were held in the treated soil for 1 week. First-instar grubs were held for 3 days in one test and for 1 week in a second test.

Pupae were exposed to the fumigant by placing five mature third-instar grubs in each pot and applying the pour-on treatment after a majority pupated. Treated pots were held for 1 week.

Susceptibility of adults was determined by placing five beetles of each sex in each pot, placing window screen across the top of the pots to prevent the escape of beetles, then applying the necessary amount of the fumigant. Beetles were recovered in 3 days.

The results, given in table 15, showed that the pour-on treatment was effective against all stages. A few eggs hatched but the young grubs died within 2 weeks. All first-instar grubs died during a 1-week exposure period, and 19 percent that survived a 3-day exposure period died within 1 week. The few pupae that survived the 1-week exposure period died within 2 weeks. All adults died within 4 days regardless of sex.

Injection of Soils

Eggs were treated by preparing soil balls identically to those used for immersion treatments and injecting the fumigant immediately after their preparation. They were held for 3 days.

Fifty recently hatched first-instar grubs were placed in a cheesecloth bag with soil, the bag surrounded with window screen for protection, and one of these placed in the center of each artificial soil ball. They were immediately injected with the fumigant and held for 1 week. Because of the rapid death and decomposition of these small grubs only nine were recovered.

Pupae were treated by infesting spirea soil balls with 25 mature third-instar grubs, holding them until a majority had pupated, then injecting the fumigant. These were held for 1 week before recovering the pupae.

Susceptibility of adults was determined by placing 25 beetles of each sex in each artificial soil ball, injecting the soil with the fumigant, and recovering the beetles in 3 days.

Injection of ethylene dibromide was highly effective against all stages, as shown in table 15. All but two of the treated eggs were recovered but none of them hatched. All the first-instar grubs and pupae exposed to the fumigant were killed during the 1-week exposure period. At the end of the 3-day exposure period 99 percent of the beetles were dead and the remaining few died within 1 week.

TOLERANCE OF PLANTS TO ETHYLENE DIBROMIDE

Balled and Burlapped Evergreens

Preparation of Plants and Testing Procedures

Each cooperating nurseryman prepared the balled and burlapped evergreens according to his usual practice. Since the plants were dug during late spring after the regular shipping season was nearly over, some of the species had produced considerable new growth. To simulate commercial practice, most of the plants were held for 1 to 4 days before use, either on the loading platform or in the packing shed. All plants were treated between May 22 and June 7. Four or five plants of each species were used in each treatment at each nursery, including the checks.

Formulation 431 was added to water to produce a concentration of 0.54 gram of ethylene dibromide per gallon. The soil of each plant was immersed in this solution until saturated and then allowed to drain. Other soil balls were injected with formulation 434. The dosage of 0.8 gram of ethylene dibromide per cubic foot of soil was introduced following the injection pattern advised by Fleming et al. (1).

The plants were held for 1 week before planting to approximate the time normally passing from digging to planting. They were then planted at each nursery site according to accepted practices.

Plant Reactions

None of the plants showed any evidence of injury during the 1-week preplanting period. Observations were made 4 days later, 1 week later, and at weekly intervals for 4-5 weeks. The final observations were made 3 months after treatment. Plants were considered tolerant to ethylene dibromide when the treated individuals appeared similar to the untreated plants. They were considered susceptible when they were retarded in growth, discolored, or killed. Definite reactions were apparent on the more susceptible plants during the first week. Injury first appeared as wilting and yellowing of the leaves. The most susceptible plants were dead within 4 weeks. Less susceptible species were not killed but remained in an unhealthy, stunted condition. Definite differences occurred in the reactions of the various plants to ethylene dibromide. Also, there were differences in reactions of the same species at the different nurseries.

The reactions of the plants to ethylene dibromide were as follows:

Species tolerating both methods of application:
Juniperus virginiana 'Canaertii'
Thuja occidentalis 'Douglasii pyramidalis'
T. occidentalis 'Globosa'
T. occidentalis L.
Tsuga canadensis (L.) Carr

Species tolerating immersion, injured by injection:
Juniperus communis 'Hibernica'
Pinus mugo var. mughus Zenari
Pseudotsuga menziesii (Mirb.) Franco
Taxus cuspidata Sieb. & Zucc.
T. media 'Hicksii' Rehd.

Species susceptible to both methods of application:
Juniperus chinensis 'Pfitzeriana'
Taxus media 'Brownii'
T. media 'Hunnewelliana' (= 'Hatfieldii'?)

The reaction of Picea abies Karst., P. glauca var. densata Bailey, and P. pungens var. glauca Regel could not be determined because untreated plants were unhealthy. It appeared that the lateness of the treatments in the spring contributed significantly to the injury. Those plants that had produced the most new growth, specifically the species of Taxus, were most injured. Species that had not produced any new growth, as Thuja, produced no reactions to the treatments.

Potted Plants

Testing Procedures

Potted plants, including about 60 different varieties at 7 different establishments, were treated with ethylene dibromide by several methods of application. Most of the plants were in vigorous growing condition and many of the flowers were in bloom. The following five treatments were used:

1. Application of 0.8 gram of ethylene dibromide per cubic foot of soil in a volume of water equivalent to one-fifth the soil being treated.
2. Application of 0.8 gram in a volume of water equivalent to one-tenth the soil.
3. Immersion of the pots in an aqueous solution containing 0.54 gram of fumigant per gallon.
4. Injection of miscible ethylene dibromide at the rate of 0.4 gram per cubic foot of soil.
5. Injection at the rate of 0.8 gram per cubic foot.

During 1956, 10 plants of each species at each nursery were exposed to the fumigant (treatment No. 1). During 1957, only five plants were included in each treatment at each location, but each species was treated by either four or five methods, with an equal number of untreated plants held as checks. All the plants were selected from the regular stock grown for sale. After treatment they were held on the greenhouse benches at each establishment and cared for by the growers according to their usual practice. In addition to greenhouse plants, roses and hardy chrysanthemums, grown in 7-inch asphalt paper pots and held outdoors, were treated. Observations were first made within $\frac{1}{2}$ week and at weekly intervals for 4 weeks.

Plant Reactions

Reactions of susceptible species were evident within a week. Wilting and yellowing of leaves were the first symptoms. As the injury progressed, these leaves died and fell from the plants. In contrast to the evergreens, outright death of plants was rare; most of them recovered eventually and produced new growth. In spite of recovery plants were not salable because they were misshapen. In general, plants that reacted to ethylene dibromide when treated by one method of application reacted similarly to all methods.

Normal, healthy plants of the following species and varieties were not injured by ethylene dibromide:

Ageratum, 'Riverside'
Aralia elegantissima = Dizygotheca elegantissima Vig. & Guill.
Azalea (see Rhododendron)
Begonia semperflorens 'Westport Beauty'
Buxus microphylla var. japonica Rehd. & Wils.
Cissus, 'Maple Leaf'
'Maple Queen'
'Summer'
'Variegated'
Coleus sp.
Collinia elegans Liebm.
Crassula arborescens Willd.
Dracaena godseffiana Sander
D. indivisa (=Cordyline indivisa Kunth)
D. sanderiana Sander
Euonymus, 'Silver'
E. japonicus 'King Midas'
Hedera canariensis 'Variegata' Schulze
Hydrangea macrophylla 'Hamburg'
'Kunert'
'Merveilla'
'Strafford'

Nephrolepis exaltata var. bostoniensis Davenport
Pelargonium domesticum 'Lady Washington'
Peperomia hederifolia 'Watermelon'
Petunia, 'Ruffled Hybrid'
Philodendron cordatum Kunth
Pilea cadierei Gagnep. & Guill.
Rhododendron, Belgian Indian hybrids, 'Hexe de Saffelaere'
'Pink Pearl'
'Pride of Sommerville'
'Triumphe'
Rhododendron, Kurume hybrids, 'Coral Belle'
'Snow'
Rhododendron, Pericat hybrids, 'Madam Pericat'
'Pink Pericat'
'Sweetheart'
'Twenty Grand'
Rosa, [Cl.] 'Gladiator'
[F.] 'Goldilocks'
[H. T.] 'Kathryn T. Marshall'
Salvia, 'St. John's Fire'
Sansevieria, 'Tom Thumb'
S. trifasciata var. laurentii N. E. Br.
Saxifraga sarmentosa 'Magic Carpet'
Syngonium podophyllum 'Emerald Gem'
Tagetes sp.
Vinca sp.

The following species and varieties of plants were normally tolerant to ethylene dibromide, but were susceptible when in poor condition brought about by overwatering:

Ageratum, 'Blue Mink'
Coleus sp.
Petunia, 'Erfurt Giant'
Tagetes, 'Naughty Marietta'

These plants showed evidence of damping-off. They were more readily injured by the immersion or pour-on treatments than by the injection treatments.

Normal, healthy plants of the following species and varieties were always susceptible to ethylene dibromide:

Pelargonium, 'Anderson's Wonder'
'Ann Vincent'
'Hassey Scarlet'
'Olympic'
'Penney'

Pelargonium, 'Salmon Ideal'
'White'
Senecio cineraria 'Fern Leaf'

Hardy chrysanthemums growing in the greenhouse were injured by the pour-on treatment, but plants grown outdoors and potted during the fall in 7-inch asphalt paper pots tolerated ethylene dibromide. The varieties treated were Avalanche, Cardinal Spoon, Rose Spoon, and Sunapee.

Vegetable Plants

Vegetables grown as transplanting stock are generally sold in trays or plant bands filled with soil. Such plants grown in the chafer-infested areas could become subject to the quarantine against this insect. Therefore, the reaction of the more common vegetables grown in this manner to ethylene dibromide was determined.

Treating Procedures

Tomato, pepper, eggplant, cabbage, and cauliflower plants were grown in greenhouse flats until they approached transplanting size. An aqueous solution of ethylene dibromide was applied to the soil surface so that 0.8 or 1.6 grams of the fumigant per cubic foot of soil was distributed in a volume of water equal to one-fifth or one-tenth the volume of the soil being treated. Tomato, pepper, eggplant, cucumber, muskmelon, summer squash, and watermelon were grown in individual plant bands until they approached transplanting size. Then they were immersed in an aqueous solution containing 0.4 or 0.54 gram of ethylene dibromide per gallon. Bare-rooted cabbage and cauliflower transplants were immersed in water containing 2.4 grams of ethylene dibromide per gallon. These plants were observed at half-week intervals for 3 weeks, until they grew too large for maintenance in flats and plant bands.

Plant Reactions

The following varieties of vegetables were tolerant to ethylene dibromide by all methods to which they were exposed:

Cabbage var. Seneca Danish Ballhead
Special Golden Acre
Cauliflower var. Snowball Perfected
Cucumber var. Niagra
York State Pickling
Muskmelon var. Delicious 51
Iroquois
Pepper var. Early California Wonder
Penwonder

Summer Squash var. Seneca Zucchini Hybrid
Yankee Hybrid
Watermelon var. Golden Honey Cream
Rhode Island Red

Tomatoes (var. Valiant, Red Jacket, and Long Red) were mildly susceptible to ethylene dibromide. The application of 1.6 grams of fumigant per cubic foot of soil caused wilting of plants and death of the actively growing root tips. Eggplant (var. Black Beauty) was highly susceptible. The leaves wilted within a half week after treating and the plants were dead within a week.

SUMMARY

Ethylene dibromide was applied to soils of balled and burlapped and potted plants to control third-instar grubs of the European chafer. The three methods of application and the concentrations of the fumigant required to eliminate the grubs were as follows: (1) Immersion of balled and burlapped soils into solutions containing 0.48 gram per gallon and potted soils into solutions containing 0.4 gram per gallon, (2) surface application of 0.8 gram per cubic foot of potted soils, and (3) injection of 0.8 gram per cubic foot of balled and burlapped soils and 0.4 gram per cubic foot of potted soils. Insecticidal action was definitely slower with the injection treatment than with the other two methods of application.

Three formulations were tested for immersion and pour-on treatments and two for injection treatments. Replacement of technical ethylene dibromide with Dowfume W-85 did not influence the effectiveness of the fumigant. Chlordane in the treating solution did not increase its insecticidal action. Immersion of soil for 10 seconds in the treating solution did not eliminate the grubs; thorough saturation was required. Applying the fumigant to the surface of Farmington loam in a volume of water equivalent to one-tenth the volume of soil being treated was equally as effective as using a volume equivalent to one-fifth that of the soil; in Canadian peat the larger volume of water was slightly superior to the smaller volume.

Soil temperature during the 1-week exposure period was of minor importance. Increase in temperature caused an increase in mortality during the exposure period but had no influence on the time required to eliminate the grubs. In immersion tests a minimum temperature of 34° F. was required during the exposure period to eliminate the grubs. In other methods of application, grubs were eliminated with temperatures down to 32° F. during the exposure period. When the fumigant was injected, subsequent freezing of the soil ball did not reduce the effectiveness of the treatment.

The influence of soil type on the effectiveness of ethylene dibromide depended on the method of application. Soil type did not influence effectiveness of the immersion treatment. With the pour-on treatment, grubs were eliminated within 4 weeks in Farmington loam, and within 6 weeks in Canadian peat. When the fumigant was

injected, grubs were eliminated in about 6 weeks in the mineral soil and up to 9 weeks in Canadian peat.

Eggs, first-instar grubs, pupae, and adults were exposed to ethylene dibromide applied by the three methods. Eggs, pupae, and adults were more susceptible than third-instar grubs and were eliminated within 2 weeks.

Balled and burlapped evergreens, potted flowers and foliage plants, and vegetables were exposed to ethylene dibromide. Definite differences in reaction by the various species to the fumigant occurred. Thuja spp. were the most tolerant evergreens. Dormant evergreens tolerated the fumigant better than vigorously growing plants. Immersion was generally safer than the injection treatment. Most potted greenhouse plants tolerated the fumigant when healthy but were injured when in poor condition. Injection appeared slightly safer than the surface-application and immersion treatments on plants in poor condition caused by overwatering. Varieties of Pelargonium (exclusive of Lady Washington type) and Senecio cineraria were always susceptible to the fumigant. Seven kinds of vegetable plants tolerated ethylene dibromide. Tomato was slightly susceptible and eggplant highly susceptible.

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Table 1.--Effect of concentration of ethylene dibromide (formulations 253 and 431) in the treating solution against third-instar European chafer grubs through immersion of balled and burlapped and potted soils

Amount of ethylene dibromide in treating solution (g./gal.)	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
Balled and burlapped Ontario loam						
0.36	99	0	90	95	95	95
.42	99	9	97	97	97	97
.48	50	4	93	100	100	100
.54	99	21	87	100	100	100
Untreated	99	4	14	14	14	19
Potted Farmington loam						
0.26	92	13	69	88	98	98
.33	137	31	85	99	100	100
.40	182	39	87	95	99	100
.47	98	31	93	99	100	100
.54	50	10	82	98	100	100
Untreated	92	2	7	7	8	15
Potted Canadian peat						
0.26	94	36	79	89	90	98
.33	134	39	84	93	97	98
.40	197	47	92	98	99	100
.47	99	21	100	100	100	100
.54	50	0	94	100	100	100
Untreated	95	6	10	12	18	19

Table 2.--Efficiency of ethylene dibromide at the rate of 0.54 gram per gallon with and without chlordane in eliminating third-instar European chafer grubs through immersion of balled and burlapped soil

Soil temperature during exposure period (° F.)	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
Formulation 253, with chlordane						
50	123	85	100	100	100	100
44	117	50	100	100	100	100
39	123	49	100	100	100	100
36	245	20	100	100	100	100
36	243	2	94	100	100	100
35	127	16	98	99	99	100
Formulation IDM-401, without chlordane						
50	125	93	100	100	100	100
44	125	44	100	100	100	100
39	121	41	100	100	100	100
36	123	10	100	100	100	100
36	124	9	98	99	100	100
35	124	6	88	97	97	98
Untreated						
Mean, all temp.	505	7	11	11	14	15

Table 3.--Influence of source of ethylene dibromide (formulation) and temperature during the 1-week exposure period on the effectiveness of ethylene dibromide at 0.54 gram per gallon against third-instar European chafer grubs through immersion of balled and burlapped soil

Soil temperature during exposure period (° F.)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
		Formulation 253			
40	125	34	100	100	100
50	124	51	100	100	100
60	124	88	98	99	100
70	123	100	100	100	100
		Formulation 431			
40	124	6	100	100	100
50	123	66	100	100	100
60	120	93	100	100	100
70	110	99	100	100	100
		Untreated			
Mean, all temp.	178	10	10	10	10

Table 4.--Influence of period of immersion of balled and burlapped soil in a concentration of 0.54 gram of ethylene dibromide per gallon on mortality of third-instar European chafer grubs

Period of immersion	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
10 seconds	123	4	88	97	97	97
	120	2	69	89	89	96
To saturation	123	1	99	100	100	100
	118	4	89	100	100	100
Untreated	100	4	11	11	11	12

Table 5.--Influence of soil type and temperature on the effectiveness of ethylene dibromide against third-instar European chafer grubs through immersion of balled and burlapped and potted soils

Soil temperature during exposure period (° F.)	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
Balled and burlapped Farmington loam in 0.54 g. EDB/gal.						
32	125	5	58	71	81	87
45	123	11	97	98	100	100
60	110	96	98	99	100	100
70	117	94	100	100	100	100
Untreated	95	20	20	24	24	24
Balled and burlapped Canadian peat in 0.54 g. EDB/gal.						
32	125	5	77	85	90	93
45	122	5	97	99	100	100
60	117	95	100	100	100	100
70	118	95	100	100	100	100
Untreated	98	10	10	10	10	12
Potted Farmington loam in 0.4 g. EDB/gal.						
34	50	6	90	94	100	100
40	50	10	84	94	100	100
50	50	20	92	94	100	100
60	36	75	94	100	100	100
70	46	50	80	93	98	100
Untreated, mean	115	0	4	5	5	7
Potted Canadian peat in 0.4 g. EDB/gal.						
34	50	4	100	100	100	100
40	50	10	94	100	100	100
50	48	48	96	100	100	100
60	49	53	80	91	96	100
70	48	75	98	100	100	100
Untreated, mean	115	0	4	6	9	9

Table 6.--Influence of dosage of ethylene dibromide, with and without chlordane, applied to potted soil on the mortality of third-instar European chafer grubs

Grams per cubic foot of soil		Number of grubs recovered	Percent mortality after--				
EDB	Chlordane		1 week	2 weeks	3 weeks	4 weeks	5 weeks
			Formulation 253				
0.4	0.2	79	21	59	76	83	86
.6	.3	166	17	77	97	99+	99+
.8	.4	169	28	84	99	100	100
			Formulation IDM-401				
0.4	-	78	17	47	68	70	80
.6	-	168	21	61	87	90	90
.8	-	169	30	73	99	100	100
			Untreated pots				
0	-	48	4	4	4	7	8

Table 7.--Influence of soil type and temperature during the 1-week exposure period on the effectiveness of 0.8 gram of ethylene dibromide per cubic foot applied to potted soil in eliminating third-instar European chafer grubs

Soil temperature during exposure period (° F.)	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
		Farmington loam				
40	94	8	100	100	100	100
50	91	51	100	100	100	100
60	95	98	100	100	100	100
70	94	99	100	100	100	100
		Dunkirk fine sand				
40	96	35	100	100	100	100
50	93	35	100	100	100	100
60	88	97	100	100	100	100
70	89	100	100	100	100	100
		Canadian peat				
40	95	9	98	100	100	100
50	90	26	95	98	99	99
60	85	61	89	95	100	100
70	94	79	94	99	99	99
		Untreated (mean, all soil types and temperatures)				
	328	14	14	14	14	14

Table 8.--Influence of volume of treating solution used to apply 0.8 gram ethylene dibromide per cubic foot of mineral and organic soils in pots on the mortality of third-instar European chafer grubs

Soil temperature during exposure period (° F.)	Volume of liquid added in relation to volume of soil	Number of grubs recovered	Percent mortality after--				
			1 week	2 weeks	3 weeks	4 weeks	5 weeks
		Farmington loam					
Below 40	1/5	50	0	71	94	100	100
	1/10	50	0	92	98	100	100
40-50	1/5	95	15	100	100	100	100
	1/10	101	31	100	100	100	100
60-70	1/5	90	100	100	100	100	100
	1/10	87	100	100	100	100	100
All temp., untreated		120	8	28	32	32	32
		Canadian peat					
Below 40	1/5	49	6	90	98	98	98
	1/10	50	6	66	92	98	98
40-50	1/5	98	17	99	99	99	99
	1/10	97	11	84	93	99	99
60-70	1/5	87	86	97	99	99	100
	1/10	86	47	85	91	96	98
All temp., untreated		113	4	4	4	4	4

Table 9.--Control of third-instar European chafer grubs in potted roses through application of ethylene dibromide at the rate of 0.8 gram per cubic foot

Soil temperature during exposure period (° F.)	Number of grubs recovered	Percent mortality after--				
		1 week	2 weeks	3 weeks	4 weeks	5 weeks
40-50	124	11	100	100	100	100
	113	16	97	99	99	100
35-40	125	3	95	95	95	99
	102	20	87	89	96	99
Mean, all checks	149	5	9	15	21	25

Table 10.--Influence of dosage of ethylene dibromide injected into balled and burlapped and potted soils on the mortality of third-instar European chafer grubs

Grams of ethylene dibromide per cubic foot	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	4 weeks	6 weeks
		Balled and burlapped Ontario loam			
0.8	101	21	85	97	99
1.2	99	35	97	100	100
1.6	98	36	80	100	100
Untreated	50	2	10	10	10
		Potted Farmington loam			
0.4	88	53	91	97	100
.6	76	76	93	100	100
.8	82	95	100	100	100
Untreated	74	3	21	40	40
		Potted Canadian peat			
0.4	84	23	65	85	91
.6	87	33	74	97	97
.8	90	45	82	97	98
Untreated	86	1	3	17	19

Table 11.--Influence of size of soil balls on the effectiveness of ethylene dibromide injected into balled and burlapped soil at the rate of 0.8 gram per cubic foot against third-instar European chafer grubs

Diameter of soil balls (inches)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	3 weeks	4 weeks
6	499	29	99	99+	100
8	501	37	91	99	99+
10	503	39	96	99+	99+
Untreated	310	4	13	17	22

Table 12.--Influence of formulation of ethylene dibromide injected into balled and burlapped soil at the rate of 0.8 gram per cubic foot and of soil temperature during the exposure period on the mortality of third-instar European chafer grubs

Soil temperature during exposure period (°F.)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	4 weeks	6 weeks
2.5% miscible ethylene dibromide					
40	122	22	95	100	100
50	120	59	95	98	100
60	119	76	85	93	99
70	124	87	95	100	100
Formulation 434					
40	123	2	100	100	100
50	114	42	95	100	100
60	119	77	88	99	99
70	113	78	95	99	100
Untreated					
Mean, all temp.	388	17	17	17	17

Table 13.--Influence of soil type and temperature on the effectiveness of ethylene dibromide injected into balled and burlapped and potted soils against third-instar European chafer grubs

Soil temperature during exposure period (°F.)	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	4 weeks	6 weeks
Injection of 0.8 g./cu. ft. into balled and burlapped Farmington loam					
32	125	16	99	100	100
40	125	23	99	100	100
50	123	51	93	97	99
60	115	76	90	97	100
70	125	83	92	99	99
All temp., untreated	122	1	7	20	20
Injection of 0.8 g./cu. ft. into balled and burlapped Canadian peat					
32	125	8	96	100	100
40	123	5	91	98	99
50	124	43	93	98	100
60	117	60	78	95	98
70	125	76	88	94	95
All temp., untreated	123	2	4	18	32
Injection of 0.4 g./cu. ft. into potted Farmington loam					
33	50	2	98	98	98
40	50	2	95	100	100
50	49	14	90	97	97
60	45	64	97	100	100
70	50	46	68	100	100
All temp., untreated	116	6	9	11	15
Injection of 0.4 g./cu. ft. into potted Canadian peat					
33	50	2	98	100	100
40	50	8	75	100	100
50	48	9	77	93	100
60	44	26	57	87	98
70	48	27	67	83	92
All temp., untreated	125	2	6	6	7

Table 14.--Influence of freezing of soil balls after injection of ethylene dibromide at the rate of 0.8 gram per cubic foot on its effectiveness against third-instar European chafer grubs

Condition of soil balls	Number of grubs recovered	Percent mortality after--			
		1 week	2 weeks	4 weeks	6 weeks
Treated, not frozen	124	54	98	99	100
Treated, frozen	125	42	93	97	100
Untreated, not frozen	49	0	2	6	8
Untreated, frozen	50	56	60	62	68

Table 15.--Susceptibility of several stages of the European chafer to ethylene dibromide

Stage of chafer	Number of individuals recovered		Percent mortality within 1 week[1/]	
	Treated	Untreated	Treated	Untreated
Immersion of soil in 0.54 gram of EDB per gallon of water				
Eggs	247[2/]	50	100	64
First-instar grubs	2	33	100	3
Pupae	213	32	99[3/]	7
Pour-on of 0.8 gram EDB per cu. ft., potted soil				
Eggs	343	138	99	10
First-instar grubs	413[4/]	239	100	5
Pupae	84	23	98	14
Adults	100	30	100	37
Injection of 0.8 gram EDB per cu. ft., balled and burlapped soil				
Eggs	248[2/]	50	100	14
First-instar grubs	9	31	100	38
Pupae	49	22	100	23
Adults	154	50	100	30

1/ 100-percent mortality in all treatments within 2 weeks.

2/ Initial 250 eggs held for hatching; small grubs decayed too rapidly for larger recoveries with a 1-week exposure.

3/ Pupae exposed to additional concentrations of 0.27, 0.41, and 1.08 gram per gallon.

4/ Initial 1,000 eggs held for hatching; recovery was from 3-day exposure.

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